In the Specification

Line and page numbers with these instructions are the Applicants best judgement as to location based on the filing of previous amendments to the Specification and the line and page numbers in the original PCT application. It is possible, however, that the actual numbers may be slightly different than what is cited here.

Please add the following new paragraph after the paragraph on page 8 (that was requested to be added to the Specification in the Amendment/Response of 6/29/04) that begins with the words "Conventional linkage study techniques, including conventional association-based studies and linkage analysis, are essentially one-dimensional":

Versions of the invention are based on the principle that the power of association-based linkage tests is increased as the frequencies of a bi-allelic trait-causing polymorphism allele and positively associated bi-allelic marker allele become similar in magnitude. Systematically covering a two-dimensional CL-F region with markers increases the power of association based linkage studies to detect evidence for linkage. In general the closer (the smaller the covering distance) in terms of either frequency distance, chromosomal location distance, or both, the greater the power of a study that uses the markers to detect evidence for linkage. Versions of the invention also use a new, two-dimensional concept of "closeness" for association-based linkage studies (some examples are: versions wherein the sought trait-causing polymorphism is unknown, and versions that use N-coverings, wherein $N \geq 2$ to increase the likelihood of one or more markers being in linkage disequilibrium, especially strong positive linkage disequilibrium, with the sought trait-causing polymorphism).

Please add the following six new paragraphs (in the order given below) on the page after Table 3 that was requested to be added to the Specification in the previous Amendment/Response of June 29, 2004 but before the heading "**Industrial Applicability**" now on page 52 that was formerly on page 47 line 30 of the PCT application:

The equations for P_t, P_s and H/F can be used to compare the power of χ^2_{tdt} and χ^2_{asp}. It is assumed that the two tests consider markers that are tightly linked ($\theta = 0$) to bi-allelic disease loci with additive mode of inheritance ($\beta = (\alpha + \gamma)/2$) and for which the α:γ penetrance ratio is r = 2, r = 4 or r = 10. Penetrance ratios of r = 2, 4 and 10 were chosen as being somewhat representative of the entire genetic parameter space since the inventor has found that P_t and P_s increase rapidly as r increases from 2 to 6 with smaller, asymptotic increases in P_t and P_s for r > 10. Furthermore, additive mode of inheritance may also be regarded as being somewhat representative since results from other modes of inheritance do not, in general, substantially differ from results presented here. In the Tables 1, 2 and 3 above, χ^2_{tdt} and χ^2_{asp} are compared when both tests consider the same bi-allelic marker, or when χ^2_{asp} considers a fully informative marker and χ^2_{tdt} evaluates a nearby bi-allelic marker. Such single test comparisons would be occasioned by: (a) TDT and ASP analysis of a marker that gave 'suggestive' evidence of linkage and disease-association in other families or in comparisons of allele frequencies in cases and unrelated controls; or (b) TDT and ASP analysis of markers near a candidate gene suspected of increasing disease susceptibility. (See page 164 of the inventor's published paper Annals of Human Genetics, 1998, vol. 62, pp. 159-179, abbreviated as AHG 98 herein.) The algebraic form of P_t* for singletons (as opposed to ASPs) is similar to P_t and the values of P_t* and P_t are identical for the Risch & Merikangas model and are similar (though not identical) for many other genetic models (see AHG 98 pp. 168 and 169). The power calculations in Tables 1, 2 and 3 and the mean χ^2_{tdt} calculations above use association values between marker and polymorphism allele pairs that are known (i.e. known association data values).

The following Background information is found in the inventor's paper (AHG 98) and is used in the power and mean χ^2_{tdt} calculations given above. The paper compares the transmission/disequilibrium test (TDT) and affected sib pair (ASP) test under a general algebraic model describing a bi-allelic disease locus. Assuming linkage to a bi-allelic marker, two binomial probabilities are derived, one for parental allele 'transmission' *(P_t)* which determines the magnitude of the TDT χ^2 statistic (χ^2_{tdt}), and a second for identity-by-descent (ibd) marker allele 'sharing' *(P_s)* which determines the magnitude of the ASP test statistic (χ^2_{asp}). A general framework for determining the power of the TDT and ASP test is presented based on expressions for *P_t*, *P_s* and the proportion *(H/F)* of ascertained parents who are informative at the marker. (See Abstract AHG 98 p. 159)

The χ^2_{tdt} statistic for detecting linkage by the TDT is $\chi^2_{tdt} = (n_a - n_b)^2 / (n_a + n_b) = (n_a - n_b)^2 / n_{tdt}$ where n_a and n_b are the number of instances in which an A/B parent transmitted allele A or B, respectively to an individual affected offspring; and thus $n_a + n_b = n_{tdt}$ is the sample size for χ^2_{tdt} (see AJHG 98 p. 161). Based on sample size (n_{asp}, n_{tdt}) and binomial probability (P_s, P_t), two binomial distributions are generated which can be used to calculate the power of χ^2_{asp} and χ^2_{tdt} as described in Appendix II of the paper. Specifically, the power of χ^2_{asp} or the probability that $\chi^2_{asp} > L$ (a significance cutpoint) is equal to the portion of the binomial distribution based on P_s for which $n_s > n_{asp}/2 + (\sqrt{(n_{asp} L)})/2$. Similarly, if marker allele A is associated with disease, the power of χ^2_{tdt} is estimated by the portion of the binomial distribution based on P_t (below) for which $n_a > n_{tdt}/2 + (\sqrt{(n_{tdt} L)})/2$. The expression for the binomial distribution based on P_t is:

$$\frac{n_{tdt}!}{n_a! n_b!} P_t^{n_a} (1 - P_t)^{n_b}$$

Thus, standard tables giving the normal approximation to the binomial distribution (Pearson & Hartley, 1954, Weir, 1990) give precise power values for virtually any sample size (n_{asp}, n_{tdt}), binomial probability (P_s, P_t), and significance level (see AHG 98 p. 164).

The TDT association studies or tests in the Risch & Merikangas analysis are referred to in the Background of this patent application (e.g., see Risch & Merikangas analysis and the Muller-Myshok and Abel criticism/letter pages 5 and 6 of the Background). In the Risch & Merikangas analysis, the TDT was assumed to test the disease locus itself or a perfectly associated bi-allelic marker, i.e. association with $m = p$ and $\delta = \delta_{max}$ (see AHG 98 pp. 166, 169 and Background pp. 5 and 6; perfectly associated markers are also discussed on pp. 178, 179 of AHG 98 and p. 45 lines 28 to 31 of the PCT application). For each TDT association study or association test in the Risch & Merikangas analysis a marker allele with known association to each possible disease-causing polymorphism allele (in each such association study or test) is tested; and the known association values or data is $m = p$ and $\delta = \delta_{max}$ for each such possible disease-causing polymorphism. The inventor's work has extended the analysis of Risch & Merikangas from the optimal situation of a perfectly associated marker for each possible disease-causing polymorphism (in each association study or test) to include the more common, less optimal situations in which $m \neq p$ and, or $\delta \neq \delta_{max}$ for tested marker-possible disease-causing polymorphism pairs (see the pages 5 and 6 of the Background of this patent application and information above from AHG 98 pp. 166 to 171).

The extension of the Risch & Merikangas analysis to include these more common, less optimal situations is illustrated by markers and disease-causing polymorphisms in tables in the patent application. These disease-causing polymorphisms and markers are described in the application in Table 2 (page 40 of the PCT application) and Tables 1, 2 and 3 above of AHG 98. The increased power of these markers to detect evidence for linkage to a disease-causing polymorphism is described in the application, and the increased power is also quantified and exemplified by mean χ^2_{tdt} calculations. A particular example mean χ^2_{tdt} calculation that illustrates the increased power when $m \neq p$ and, or $\delta \neq \delta_{max}$ is on p. 166 of AHG 98 and is given above. Another such example mean χ^2_{tdt} calculation in the Theory of Operation Section is also $800(H/F)(2P_t - 1)^2$ and uses $r = 5$, additive mode of inheritance and sample size of 200 hundred families with 2 affected sibs. The power calculations in all the tables above and the mean χ^2_{tdt} calculations use association values between marker and polymorphism allele pairs that are known (i.e. known association data values) in terms of known values of allele frequencies m and p and known values of δ/δ_{max} when $\delta \geq 0$ and δ/δ_{min} when $\delta < 0$. (The well-known formulas for determining δ_{max} and δ_{min} in terms of m and p are on page 178 of AHG 98.)